THE LIGHT-RECEPTIVE ORGANS OF CERTAIN BARNACLES.[1]

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Introduction.

Although brief references to the light-perceptive organs of barnacles are found throughout the literature, no really comprehensive study has been made of them since that by Pouchet and Jobert in 1876. The relationships between the "eyes" in the larval stages and those in the adult have never been fully worked out. Accordingly the object of this investigation has been a study of the various light-perceptive organs found in the larval and adult barnacles and the determination of their relationships.

Investigations relating to the light-receptors of barnacles are usually found combined either with studies of other structures of the barnacle or with studies of the eyes of Crustacea in general. Among the former type Darwin's "Monograph of the Cirripedia" (1851 and 1854) is outstanding. More recently Gruvel (1905) has published a detailed account of the morphology and taxonomy of this group. Grenacher (1879), Claus (1891), Brooks and Herrick (1892), and Demoll (1917) have studied the eyes of Crustacea. General problems in this connection have been taken up by Parson (1831), Patten (1886 and 1887), and Mark (1887).

[1] The author is indebted to Dr. J. P. Visscher of Western Reserve University for the suggestion of the problem and for assistance during the course of the investigation.

Materials and Methods.

The ivory barnacle, *Balanus eburneus,* and the rock barnacle, Balanus balanoides, were used in this study. Collections were made at Woods Hole, Mass., during the spring and summer of 1926. Living forms were observed at this time and material was preserved for subsequent study.

Adults of *Balanus eburneus* brought to the laboratory often contained Nauplei nearly ready for hatching. Unhatched Nauplei were obtained by removing the ovigerous lamellæ from these adults, and free swimming forms were collected after hatching. A few metanauplei and Cyprids were obtained from towings made in the vicinity.

In order to test the actual light-perceptive function of the adult, the shell was broken so that the eyes could be exposed. Specimens with the shell thus broken were tested to make sure that the light reaction was unimpaired. The eyes were then removed by a hot needle. In all cases included in the data, the eye adhered to the needle and was removed without apparent injury to the surrounding tissue.

Material was fixed in picro-acetic formol (Bouin's) or in 10 per cent. formalin, and was either stained in Ehrlich's hematoxylin or mounted unstained. Most of the study was done by means of sections which were prepared by the ordinary paraffin method and cut from 5μ to 10μ in thickness. Grenacher's technique for the removal of pigment (Lee, 1890) was used on sections of the Cyprids. In addition a modification of Cajal's silver nitrate technique as developed by Hess (1925) was used in the study of the adult eye.

The Nauplian Stage.

The formation of the median eye in Cirripedes has never been worked out. In *Balanus eburneus* the eye first appears in the unhatched Nauplius as an elongated area of reddish pigment. Certain authors, for example Darwin (1851), have believed that the nauplian eye might arise from the union of two anlagen but, at least in *Balanus eburneus,* this does not appear to be the case. A large number of specimens in which the eye was just forming were studied but in all cases it appeared as a single area with no evidence of a double origin. In succeeding stages the pigment becomes darker and the eye appears as a bilobed structure.

The morphology of the nauplian eye of Cirripedes has usually been treated incidentally in connection with studies of the complete animal and consequently its detailed structure is known in only a few forms, chiefly among the Lepadæ. The pigmented area in the anterior region of the Nauplius was noticed by the earliest authors but their descriptions mention only the general shape of the pigment cup. A fairly complete study of the light receptors in the Cirripedia was made by Pouchet and Jobert in 1876. These authors described the nauplian eye as formed of two parts each of which represented a simple eye and was composed of a pigmented body with finely granular, rose-colored pigment and small oval bodies which stained black with osmic acid and

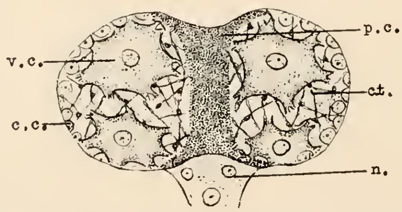

FIG. 1. Diagrammatic drawing of the median eye in the cyprid stage (longitudinal vertical section × 500). *c.c.*, cortical cells; *c.t.*, connective tissue; *n.*, nerve; *p.c.*, pigmented cells; *v.c.* visual cells.

which they thought might be designated as lenses, although the analogy was somewhat questionable. The median eye of Crustacea in general was described by Demoll (1917) as an inverted eye composed of two pigment cells forming a cup in which were the visual cells which extended proximally in a nerve fibre. These visual cells were in the nature of "Stifchensäume" and the whole eye cup was lined with fine, reflecting scales which formed the "tapetum." A cellular lens was said to be present in some forms.

When sections of fully developed Nauplei are studied the median eye (Fig. 1) can be seen lying between the so-called "cerebral ganglia." The eye at this period measures about 15μ in diameter and, due to this small size, an interpretation of its structure is very difficult. The central region is composed of heavily pigmented cells which form a bi-concave pigmented cup. In each concavity there is a non-pigmented area coated by cells with large nuclei, similar to ones which form the cortical area of the cerebral ganglia. The non-pigmented area appears to be made up of two visual cells surrounded by a small amount of connective tissue. Each area sends off a nerve which is probably formed by an extension of these visual cells as in the adult. These nerves leave the posterior region of the eye and by very short connectives enter that part of the ganglia which is slightly posterior to the eye.

A lens as described by Claparède (1863) for *Lepas* is not present in *Balanus*. The non-pigmented area composed of visual cells and connective tissue was probably mistaken for a lens by Pouchet and Jobert (1876). This description is not in accord with that given for the median eye of other Crustacea by such authors as Hesse (1901), and Demoll (1917). The eyes of the two Cirripedes studied show ganglion-like visual cells instead of the " Stabchen " or " Stiftchensäume " described by these authors. While it is difficult to determine accurately the morphology of the median eye at this period, the structure, as described, agrees fully with that observed later in development. Hanström (1927) in a recent article has described a somewhat similar structure in the larval eye of *Nymphon stromi*. Whether the lack of agreement concerning the structure is due to error on the part of the foregoing authors or whether there is a real difference in the median eye of these Cirripedia is a point which has not yet been determined.

In addition to the median eye characteristic of the Nauplius, the Metanauplius of *Balanus eburneus* has a pair of compound eyes which are generally known as the cyprid eyes since they are the most noticeable light-perceptive organs of that stage. The cyprid eyes in the Metanauplius appear on either side of the median eye near the base of each of the first pair of nauplian appendages. Their structure at this period is superficially the same as in the Cyprid but material was not available for histological verification of the appearance. The nauplian eye retains its characteristic position and appearance during the metanauplian stage.

In common with the Nauplei of other barnacles those of *Balanus* *eburneus* show definite reactions toward light. In a dish containing Nauplei these were ordinarily found congregated in the area having the greatest illumination. Occasionally a few individuals were observed which collected at a point just opposite this brightest spot. The reason for this variation has not been determined but it seems to be constant in such specimens and therefore is not the same type of reaction as that described by Groom and Loeb (1891) in which the same individuals responded differently under different conditions.

The Cyprid Stage.

The early authors had not observed the Metanauplius stage of Cirripedia and so did not always clearly recognize the distinction between the median and compound eyes. Burmeister (1834) described the median eye as a rounded, black spot which became divided and modified to form the paired, compound eyes of the Cyprid. This error persisted through several of the later works and Darwin (1854) reported Burmeister's observation although he considered it " scarcely possible that the eye of the larva of the first stage can be changed into the double eyes of the second stage." It was not until the two types of eyes were observed present at the same time that this misapprehension was fully removed.

The fate of the compound eyes at metamorphosis into the adult has been variously described. Darwin (1851) and Hesse (1874) noted that the eyes fell from their capsules during metamorphosis while Von Willemoes-Suhm (1876) stated that before metamorphosis the eyes lost their original position and might be seen only as black pigment spots which were later absorbed. A similar observation has been made by Coar [1] in an unpublished study of *Balanus balanoides* and by Hanström (1927). The author has observed the extruded eyes in slides of *Balanus amphitrite* collected by Dr. J. P. Visscher at Beaufort, North Carolina. However, the condition in *Balanus eburneus* seems to be similar to that described by Von Willemoes-Suhm and Coar. The two late cyprid stages which were observed at metamorphosis showed no compound eyes, the region of these being occupied by pigment masses which may have been the degenerating eyes.

The compound eye, as seen in total mounts, has been described

[1] Personal correspondence, 1926.

by all authors studying the Cyprid. In such preparations the eye appeared as consisting of a black pigment body and eight to ten globules or lenses surrounded by a large capsule, and is described as such by Darwin (1851), Hesse (1874), and Von Willemoes-Suhm (1876). When studied in section the compound eyes of *Balanus balanoides* (Fig. 2) are found to be situated in pockets near the bases of the antennules. In contrast to the rather unique structure of the median eye, their appearance is very similar to that of the compound eyes of other Crustacea. The eight to ten lenses described for similar forms by early authors are present and represent the same number of visual elements or ommatidia. Each ommatidium contains a cuticular lens surrounded by "corneagen cells," which are reported by Patten (1886 and 1887) to

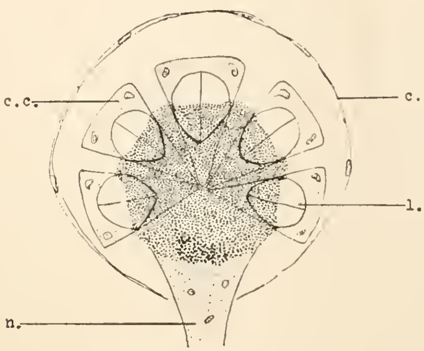

FIG. 2. Diagrammatic drawing of the compound eye in the cyprid stage (vertical section × 300). *c.*, cornea; *c.c.* corneagen cells; *l.*, lens; *n.*, nerve.

secrete this body, and other cells known as the cells of the crystalline body. The lens or crystalline body is made up of three units forming an egg-shaped structure. The proximal portion of the ommatidium contains the rhabdomes surrounded by heavily pigmented retinular cells. This region is much shorter in the compound eyes of Balanus than in most Crustacea. It was not possible to study the rhabdome in detail but it appears to be like that of other Crustacea, of reticular nature and penetrated by nerve fibrils. The retinular cells pass into the optic nerve of the compound eye. The ommatidia are surrounded by a common corneal sheath. The structure of the compound eye of *Balanus eburneus* appears to be similar, as far as could be determined from a study of total mounts.

In 1851, Bate noted that the pigmented area of the Nauplius was represented by a similar region in the Cyprid although he did not believe that the area was a light-receptor in either period. Claus (1869), in a drawing of an unknown Cyprid, pictured the persistence of the unpaired eye up to the moment of metamorphosis, although the eye is not labelled and it is probable that he did not realize the significance of his observation. Pouchet and Jobert (1876) recognized and discussed this persistence of the median eye through the cyprid stage.

The median, or nauplian, eye (Fig. 1) persists throughout the cyprid period. It has enlarged to five times its former diameter (15μ–79μ) but its structure is the same. The cells with large nuclei still coat the non-pigmented areas and because of the similarity of these nuclei to those of the regular visual cells, it is extremely difficult to determine the exact number of the latter. The bi-concave pigmented area is composed of many cells in contrast to the condition reported for *Lepas,* where only two cells are found. In each pigmented area there are two non-pigmented zones each containing two visual cells which form a nerve connection with the cerebral ganglia. These nerve connections have elongated while the amount of connective tissue surrounding the visual cells has also become greater and the reticular nature of part of it is evident in most sections.

It is difficult to say just what part the compound and median eyes play in the light perception of the Cyprid. Probably both are functional. Since the median eye is functional in the stages preceeding and following this period, as well as after the degeneration of the compound eyes, it is unlikely that it would completely lose its function at this time.

The Adult Stage.

Early observers denied the presence of an eye in adult barnacles and indeed the relatively degenerate structure and enclosing shell of the adult tended to support this view, as well as the fact that all eye structures were thought to be lost at metamorphosis, with the disappearance of the cyprid eyes. The first report on the existence of eyes in the adult barnacle was made by Leidy (1848) on *Balanus rugosus* (sp?). In 1854, Darwin substantiated Leidy's report by finding eyes present also in the adult of *Balanus tintinnabulum*. A summary of previous studies of the barnacle eye was made by Gerstaecker (1866). These were concerned chiefly with the fact of occurrence or with external appearance of the eyes. The most complete study of the barnacle eye in the adult was that made by Pouchet and Jobert in 1876.

There seems to be no published report of the origin of the eyes in the adult. Darwin (1854) pointed out that they were not developed from the eyes of the Cyprid, since the new eyes were formed at some distance from the compound, but thought they might have been formed from the nauplian eye since they occupied a similar position. Coar [1] reported that in *Balanus balanoides* the adult eyes were formed by a division of the median or nauplian eye, and the author has found that the same situation occurs in *Balanus eburneus*. The eye of the Nauplius, which has persisted throughout the cyprid stage, divides into two parts during the metamorphosis of the Cyprid into the adult. These, together with a part of the mantle which becomes modified around them, form the simple, paired eyes of the adult. In animals which have just completed metamorphosis the two eyes may be seen completely separate although still very close together (Fig. 3 *A*). They move apart during the succeeding period and, at about the fifth day, are in the position which they occupy in the adult (Fig. 3 *B*). The eyes were found to lie in the mantle between the scutum and the juncture of the rostrum (rostrum coalesced with rostro-lateral-Darwin 1851) and the lateral plates of the shell. Immediately around the eye and optic nerve the mantle lacks its usual pigmentation and for this reason the eye appears prominent. The part of the eye toward the body of the barnacle is heavily pigmented while the region toward the shell is without pigment.

Pouchet and Jobert (1876) described the eye as a rounded structure partly covered by pigment and adherent to the surrounding tissue "en arrière." They believed that this pigment func-

[1] Personal correspondence, 1926.

tioned as a choroid coat while the non-pigmented area might possibly be called a cornea. When the eye was macerated in Müller's fluid they noticed the presence of a cell which owing to its volume, granular nature, and distinct nucleolus, they felt was undoubtedly a nerve cell. The existence of a double optic nerve suggested to them the possibility that there were two such cells. However, they never observed more than one and were inclined to consider the situation analogous to that in the Lepadæ where they had found a double nerve but always a single nerve cell. The light was described as reaching the eye by traversing the tissue which united the valves and which contained no pigment in the vicinity of the eye. They found that barnacles were sensitive to light,

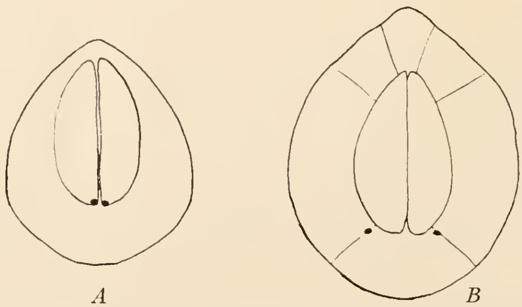

Fig. 3. Diagrammatic drawing showing adult of *Balanus eburneus*. *A*, immediately after metamorphosis; *B*, five days after metamorphosis.

but did not believe that they possessed object vision. No mention is made of the origin of the adult eye although development from the median eye is suggested by the nature of the remarks.

When sections of the eye of the adult are studied it is seen to be composed of two main divisions; an outer covering which is a modification of the mantle and an inner part which is developed from the divided nauplian eye (Fig. 4).

The outer covering is composed of irregularly shaped cells similar in appearance to those of the mantle. Pigmented cells make up about half the area of this coat and are continuous with like cells in the mantle, while non-pigmented cells form the rest of the covering and are a continuation of similar mantle cells.

Between this covering and the inner portion of the eye is a region of loose collagenous connective tissue fibers which serve to hold the inner region in position as well as to support the outer covering. In the living specimen the interstices of these fibers are filled with fluid which helps to maintain the contour of the eye.

The inner part of the eye is a sphere containing pigmented cells, collagenous fibers, reticular fibers, and visual cells with their nerves. The pigmented region forms a cap over about half of this portion of the eye and lies directly beneath the pigmented region of the outer coat. Beneath this, is an area of rather loose connective tissue fibers and cells similar to those between the outer and inner regions. The rest of the inner eye is composed of a reticulum of connective tissue fibres surrounding two large ganglion or visual cells. A slight division is observable in this

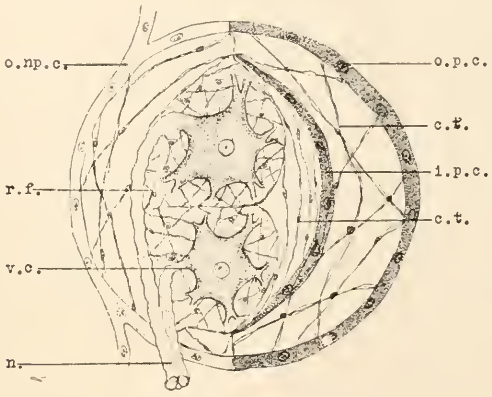

Fig. 4. Diagrammatic drawing of the simple eye of the adult (vertical section × 20). *c.t.*, connective tissue fibres; *i.p.c.*, inner pigmented cells; *n.*, nerve; *o.np.c.*, outer non-pigmented cells; *o.p.c.*, outer pigmented cells; *r.f.*, reticular fibers; *v.c.* visual cells.

reticulum, indicating the two units of nervous elements. These two visual cells send off branches which ramify throughout the reticular fibers. Each cell also gives off a nerve fiber which is surrounded by a sheath. These fibers, with their individual sheaths, become united as a double nerve enclosed within a common connective tissue coat before passing through the outer covering and continuing to the supraœsophageal ganglion as the optic nerve.

The enlargement of the nerve just prior to its entrance into the pigment mass, as described by Gruvel (1905), is not apparent in the sections and it seems probable that he has included the non-pigmented region of the outer coat as a part of the nerve. Since his work did not include studies of sections of the eye, this error is very natural.

Papers on the median eye of crustacea often describe the presence of a tapetum next to the two (or more) cells which form the eye cups. By homology the inner pigment region of the adult barnacle eye corresponds to these cells and the tapetum might therefore be either the loose connective tissue or the reticular regions of this eye. However so little uniformity is found in the use of this term by investigators that it is considered inadvisable to apply the name to any particular region of the barnacle eye.

Barnacles in their natural environment will retract their cirri when stimulated by light, and this fact is mentioned by several of the early authors. Pickering (1848) brought it forward in confirmation of Leidy's report as to the existence of eyes in the adult. Darwin (1854) observed the reaction to light in *Balanus balanoides, Balanus crenatus,* and *Chthamalus stellatus* and found that they were all sensitive to a shadow produced by passing his hand between them and the light. Gerstaecher (1866) reported experiments by Fr. Müller who found that *Balanus tintinnabulum* would react to a shadow when the body was removed and the eyes and certain muscles were left in the shell. (" dass *Balanus tintinnabulum* auf eine Beschattung mit der hand auch dann reagire, wenn er mit Zurücklassung seiner Augen an dem Manteldeckel, von diesem abgelöst werde. Ein in dieser Weise entblösstes, mit halb entrollten Ranken im Wasser liegendes Exemplar zog dieselben jedesmal schnell ein, wenn es beschattet wurde.")

In our study it was found that the adults of both *Balanus balanoides* and *Balanus eburneus* close the opercular valves if there is a sudden change in light intensity although very gradual changes do not excite the reaction. *Balanus eburneus* is more sensitive to such changes than is *Balanus balanoides.* A slight shadow may sometimes be cast upon the latter without affecting them but it was never possible to do this with *Balanus eburneus.*

Since it is shown that the adults possess some light-perceptive mechanism, it becomes necessary to find what part the so-called "eye" plays in causing this reaction. Accordingly, the light reactions were studied in twenty-five adults of *Balanus eburneus* which had been deprived of these organs and it was observed that such animals showed no reaction to light changes, however sudden or intense, although when the eyes were intact they had all withdrawn their cirri and closed the valves under similar conditions. Therefore it is concluded that these organs are the sole light-perceptors present in the adult barnacle.

The function of certain parts of the eye is problematical. Since the ganglion or visual cells send branches throughout the reticulum of connective tissue fibers immediately surrounding them, it is evident that this part serves in the transmission of the impulse. The looser collagenous fibers outside this area do not have any self-evident function. As they are not pigmented they would not prevent the passage of light and it seems probable that their function is merely that of support. The inner pigmented cells may be either protective or reflective in nature. The structure of the eye and its inverted nature lend some support to the latter possibility. The light enters the eye through the non-pigmented area of the outer coat while its entrance through the other cells is prevented by the pigment. The fluid which is found in the eye of living specimens must act as a refractive as well as a supporting medium.

Summary.

1. The development, structure, and function of the light-perceptive organs are described in the nauplian, cyprid, and adult stages of *Balanus eburneus* and *Balanus balanoides*.

2. The light-perceptive organs present in the various stages are: (*a*) nauplian—a median eye, (*b*) metanauplian—a median eye and two compound eyes, (*c*) cyprid—a median eye and two compound eyes, (*d*) adult—two simple eyes.

3. The median eye in *Balanus eburneus* originates as a single, pigmented mass in the unhatched Nauplius and persists with no change, except in size, until the metamorphosis of the Cyprid into the adult.

4. The compound eyes first appear in the metanauplian stage and remain functional throughout the cyprid stage.

5. These compound eyes are resorbed at the time of the metamorphosis of the Cyprid into the adult.

6. At the metamorphosis into the adult the median eye divides into two parts which form the simple paired eyes of the adult.

7. Each of the paired eyes in the adult is the morphological equivalent of half of the median eye plus an outer covering.

8. The simple, paired eyes are the sole light-perceptive organs of the adult.

LITERATURE CITED.

Bate, C. S.

1851 On the Development of the Cirripedia. Annals and Mag. of Nat. Hist., Second Series, Vol. 8, pp. 325–332.

Brooks, W., and F. Herrick

1892 The Embryology and Metamorphosis of the Macroura. National Acad. of Sciences, Vol. 5.

Burmeister, H.

1834 Beiträge zur Naturgeschichte der Rankenfüsser. Berlin.

Claparède, A.

1863 Beobachtungen über Anatomie und Entwickelungsgeschichte wirbelloser Thiere. Leipzig.

Claus, C.

1869 Die Cypris Aehnliche Larvae der Cirripedien und Ihre Verwandlung in das Festsitzende Thier. Schriften der Gesellschaft zur Beförderung der gesammten Naturwissenschaften zu Marburg., Bd. 9, pp. 1–17.

1891 Das Medianauge der Crustaceen. Arbeiten aus dem Zoologischen Institute der Universität Wien und der Zoologischen Station in Triest, Wien.

Darwin, C.

1851 A Monograph on the Sub-class Cirripedia with Figures of All Species. I. Lepadidæ. London.

1854 A Monograph on the Sub-class Cirripedia, II. Balanidæ. London.

Demoll, R.

1917 Die Sinnesorgane der Arthropoden, Ihre Bau und Ihre Funktion. Braunschweig.

Gerstaecker, A.

1866 Bronn's Thierreich, Arthropoda, Bd. 5, pp. 406–564. Leipzig und Heidelberg.

Grenacher, G. H.

1879 Untersuchungen über das Sehorgan der Arthropoden, insbesondere der Spinnen, Insecten und Crustaceen. Göttingen.

Groom, T., and J. Loeb

1891 Der Heliotropismus der Nauplien von *Balanus perforatus* und die periodischen Tiefenwanderungen pelagischer Tiere. Bio. Central., Bd. 10, pp. 160–177.

Gruvel, A.
1905 Monographie des Cirrhipèdes ou Thécostrăcés. Paris.
Hanström, V. B.
1927 Neue Beobachtungen über Augen und Sehzentren von Entomostraceen, Schizopoden und Pantopoden. Zool. Anzeiger, Bd. 70, pp. 236–251.
Hess, W.
1925 Photoreceptors of *Lumbricus terrestris*. Jour. Morph. and Physiol., Vol. 41, pp. 63–93.
Hesse, C. E.
1874 Description de la Série complete des Metamorphoses qui subissent, durant la periode embryonnaire, les Anatifes désignés sous le nom de Scalpel oblique ou de Scalpel vulgaire. Revue des Sciences Nat. Montpellier, T. 3, pp. 1–14, 206–212.
Hesse, R.
1901 Untersuchungen über die Organe der Lichtempfindung bei niederen Thieren. Zeit. für Wissen. Zool., Bd. 70, pp. 347–473.
Lee, A.
1890 The Microtomist's Vade-mecum. Philadelphia.
Leidy, J.
1848 Proceedings of the Academy of Nat. Sciences, Philadelphia, Vol. 4, p. 1.
Mark, E. L.
1887 Simple Eyes in Arthropods. Bull. Mus. Comp. Zoöl. Harvard College, Vol. 13, pp. 4–105.
Parson, G.
1831 An Account of the Discoveries of Müller and Others in the Organs of Vision of Insects and Crustacea. The Mag. of Nat. Hist. Edinburgh, Vol. 4, pp. 124–134, 220–234, 363–372.
Patten, W.
1886 Eyes of Molluscs and Arthropods. Mitteil. Zool. Stat. Neapel., Vol. 6, pp. 542–756.
1887 Studies on the Eyes of Arthropods. Jour. Morph., Vol. 1, pp. 193–227.
Pickering, C.
1848 Proceed of the Acad. of Nat. Sciences, Philadelphia, Vol. 1, p. 2.
Pouchet, C., and C. Jobert.
1876 Contribution a l'Histoire de la Vision chez les Cirrhipèdes. Jour. de l'Anat. et de la Physiol., T. 12, pp. 575–594.
Von Willemoes-Suhm, R.
1876 On the Development of *Lepas fascicularis* and the "Archizoea" of Cirripedia. Phil. Trans. Royal Soc. of London, London, Vol. 166, pp. 131–154.